Acute Viral Necrosis (in scallops) - Disease Card¹

By

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Pathogen information

1. Causative agent

- 1.1. Pathogen type: virus
- 1.2. Disease name and synonyms: Acute Viral Necrosis Disease (AVND)
- 1.3. Pathogen common name and synonyms: Acute Viral Necrosis Virus (AVNV)
- 1.4. Taxonomic affiliation
 - 1.4.1. Pathogen scientific name: Acute Viral Necrosis Virus (AVNV)

1.4.2. Phylum, Class, Family etc.: Unknown so far

1.5. Description of the pathogen: AVNV is an enveloped, spherical particle (130 to 170 nm in diameter) with spike-like protrusions on the surface and an inner nucleocapsid (90 to 130 nm in diameter), assembly and release of virions occurred in the virogenic matrix located in membranous cytoplasmic vesicles.

1.6. Authority:

- He, G.Z., Wang, X.H., Li, Y., Wang, C.M., Ai, H.X., Song, W.B., 2003. Infection status of the acute viral necrobiotic disease virus in different organs of scallop *Chlamys farreriv*, (Chinese J.) *High Technology* 13, 93-96.
- Song, W.B., Wang, C.M., Wang, X.H., Li, Y., 2001. New research progress on massive mortality of cultured scallop *Chlamys farreri*, (Chinese J.) *Marine Sciences* 25, 23-27.
- Wang, C.M., Wang, X.H., Song, X.L., Huang, J., Song, W.B., 2002. Purification and ultrastructure of a spherical virus in cultured scallop *Chlamys farreri*. J. Fish. China 26, 180-184.
- 1.7. Pathogen environment: sea water

2. Modes of transmission

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- 2.1. Routes of transmission: Unknown so far
- 2.2. Life cycle: Replication in the cytoplasm
- 2.3. Associated factors: Temperature at 25-27°C
- 2.4. Additional comments: none

3. Host range

- 3.1. Host type: Marine shellfish
- 3.2. Host scientific names: Chlamys farreri
- 3.3. Other known or suspected hosts: Unknown so far
- 3.4. Affected life stage: the 2-year old scallops
- 3.5. Additional comments: none

4. Geographic distribution

- 4.1. Region: North Coast of China (Shandong and Liaoning Province)
- 4.2. Country: China
- 4.3. Additional comments: none

Disease information

1. Clinical signs and case description

- 1.1. Host tissues and infected organs: Mantle, gill, kidney, intestine and digestive gland and nerves tissues.
- 1.2. Gross observations and macroscopic lesions: (1) The diseased scallop usually had slow reaction to an artificial disturbance, slow muscle contraction when closing the valves, and detachment from the valves as adductor muscle fibres broke down; (2) increased mucus and soiled material in the mantle chamber, mantle shrinkage and blemished ocelli; (3) digestive gland enlargement, and empty digestive tract. The kidney was easily detached from the adjacent tissues. However, there were no obvious inflammatory lesions on the surface of these soft tissues; (4) death occurred 2—3 days after the appearance of gross signs of disease. Severe mortality usually occurred in August at peak annual water temperature, with cumulative mortality of ~90%.
- 1.3. Microscopic lesions and tissue abnormality: Histopathological observations on moribund scallops showed lesions in gills, mantle, kidney, digestive tract and digestive gland, but not the adductor muscle or gonads. Severe multiple infections caused disordered and necrotic

architecture. Indistinct cytoplasmic eosinophilic inclusions were observed in infected tissues. Cellular changes were nuclear hypertrophy, chromatin margination, pyknosis, karyorrhexis and, lastly cellular necrosis that often left an empty space amongst the tissue lesions. The epithelium of gills, digestive tract and mantle sloughed off, there was reduction in the number of tubules in the kidney and digestive gland. Significant bacterial infection was not observed.

- 1.4. OIE status: not listed.
- 2. Social and economic significance: AVND causes significant damage to the *Chlamys farreri*. Heavy mortalities of *Chlamys farreri* in North China cause significant economic loss and affect the development of scallop industry.
- 3. Zoonotic importance: none

4. Diagnostic methods

- 4.1. Screening methods
 - 4.1.1. Level I: none
 - 4.1.2. Level II: ELISA and immunofluorescence

Indirect ELISA and immunofluorescence technique was used to detect AVNV with the polyclonal antibody and monoclonal antibody. (Fu et al., 2005; Li et al., 2003; Wang et al., 2003)

4.1.3. Level III: PCR, Real-time PCR and LAMP

PCR is a method used to amplify AVNV DNA. The primer sequence is 5'-GGA TGA TTT GCC ACT GAC GC-3' and 5'-GGC CAA GAC ATT GAC GGA AT-3' with amplified product size of 310bp.

Real-time PCR based on Taqman probe has developed for detecting and monitoring AVNV. The primer and probe sequence are c-f: 5'-AGC CTT TTA CAG AAT TTT GCA CCT T-3' and c-r: 5'-TGT CGC ATG TTA ACC TCG TCT G-3' and Taqman probe: 5'-FAM-AGC CAT CAC ATC AGC CAG CAA CGA CT-TAMRA-3' with amplified product size of 90bp.

LAMP (loop-mediated isothermal amplification) is intended to amplify DNA AVNV. Four primers able to recognize respectively six or eight sequences were used.

4.2. Presumptive methods

4.2.1 Level I: Gross observations

The scallop could die within about one week in case of the appearance of signs of the disease, during hotter summer periods when the temperature rearing at 24.5-26.7 °C.

4.2.2. Level II: Histopathology

Moribund *Chlamys farreri* showed multifocal areas of abnormalities in the connective tissue and cuticular epithelium of gills of the mantle, gill, kidney, intestine and digestive gland. These focal lesions included obviously necrotic cells and cells with hypertrophied nuclei, large vacuoles and sloughing of epithelial cells.

4.2.3. Level III: Virology

AVNV is a sphere-shaped, enveloped DNA virus which has an external bilaminal envelope with a size of 130 to 170 nm in diameter.

- 4.3. Confirmatory methods
 - 4.3.1. Level I: none
 - 4.3.2. Level II: none
 - 4.3.3. Level III: PCR, Real-time PCR and LAMP
- 5. Control methods: Because AVND is domesticated completely and PCR, real-time PCR and LAMP technique are available, brood stock and seed screening should be strongly encouraged. The brood stock or seed tested positive for AVNV must be discarded with proper methods. Usual sanitation and control protocols for viral infections are recommended.

6. Selected references:

- Fu, C.L., Song, W.B., Li, Y., 2005. Monoclonal antibodies developed for detection of an epizootic virus associated with mass mortalities of cultured scallop *Chlamys farreri*. *Dis. Aquat. Org.* 65, 17-22.
- Li, Y., He, G.Z., Wang, X.H., Wang, C.M., Song, W.B.,2003. Detection of Acute Virus Necrobiotic Disease Virus (AVND Virus) in *Chlamys farreri* using ELISA Technique. (Chinese J.) *High Technology* 13, 90-92.
- Song, W.B., Wang, C.M., Wang, X.H., Li, Y., 2001. New research progress on massive mortality of cultured scallop *Chlamys farreri*, (Chinese J.) *Marine Sciences* 25, 23-27.
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Wang, X.H., He, G.Z., Li Y., Wang, C.M., Song, W.B., 2003. Preparation of polyclonal antibody of

AVND virus and analysis by ELISA technique. (Chinese J.) High Technology 13, 84-88.

Wang, Y.T., Xiang, J.H., 1999. Studies on causation of the mass mortality of *Chlamys farreri*. (Chinese J.) *Oceanol. Limnol. Sin.* 30,770–774